Transition Metal Complexes of Aminobenzothiazole–Ketone Schiff Bases

SYNOPSIS

SUBMITTED TO

(Faculty of Sciences)

UNIVERSITY DEPARTMENT OF CHEMISTRYB. N. MANDAL

UNIVERSITY, MADHEPURA

SUBMITTED BY

NAME:- Nilesh Kumar

COURSE WORK EXAM ROLL NO.: CHE-03/23

CONTACT NO:- 7546959353

Registration No. and Year: 39/2025

Synopsis of Research Work for Ph.D. Degree in the Faculty of Science(Chemistry)

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a) Purpose and Need of the Study

Schiff bases are versatile organic ligands characterized by an azomethine (>C=N–) linkage, typically formed by condensing primary amines with carbonyl compounds. They have played a significant role in coordination chemistry as they readily coordinate to metal ions via the imine nitrogen (and other donor sites if available), forming stable chelate complexes. A wide variety of Schiff base metal complexes have been explored due to their broad utility in both chemical and biological applications. For instance, many transition metal–Schiff base complexes exhibit notable pharmacological activities (antibacterial, antifungal, anti-inflammatory, etc.) and even serve as catalysts or dyes in industrial processes. These attributes, combined with the relative ease of synthesizing Schiff bases, make them attractive targets for developing new inorganic complexes with potential functional properties.

Benzothiazole derivatives, in particular, are prominent heterocyclic compounds known for diverse bioactive properties. The benzothiazole moiety (as in 2-aminobenzothiazole) is found in various medicinal agents and has been associated with **antimicrobial, antitumor, antioxidant, and other pharmacological effects**. Incorporating a benzothiazole unit into a Schiff base ligand is expected to impart or enhance biological activity, and indeed prior studies on benzothiazole-derived Schiff bases have demonstrated potent **antibacterial, antifungal, anticancer, and cytotoxic** activities. However, most reported benzothiazole Schiff bases in literature have been obtained from aromatic aldehydes (e.g. benzaldehyde derivatives) rather than ketones. There is a clear need to explore Schiff bases formed from 2- aminobenzothiazole and ketone precursors, as these may yield novel ligands with different steric/electronic characteristics and potentially unique coordination behavior. By investigating transition metal complexes of such aminobenzothiazole–ketone Schiff bases, the proposed study aims to fill a research

gap in coordination chemistry. New insights can be gained into how the additional substitution (from ketone-derived imines) influences the structure, stability and properties of the resulting metal complexes. Ultimately, these complexes could exhibit enhanced functional properties — for example, improved antimicrobial or anticancer efficacy compared to the free ligands or useful catalytic/optical behaviors — thereby broadening the applications of Schiff base coordination compounds in medicinal and materials chemistry.

b) Details of the Project (Chapterization)

Chapter 1: Introduction – This chapter will provide a general introduction and background for the research. It will cover the fundamental concepts of Schiff base ligands and their importance in inorganic chemistry. Key historical and theoretical aspects (Hugo Schiff's discovery, general formula and formation mechanism of Schiff bases) will be mentioned to set the context. The chapter will then discuss the versatility of Schiff bases as chelating ligands and their well-known ability to form stable complexes with transition metals. Importantly, it will review the broad spectrum of applications of Schiff base complexes – ranging from biological roles (e.g. as antimicrobial, antimalarial, antiviral agents) to industrial uses (e.g. as catalysts, dyes and polymer additives) – to underscore their significance. The unique features of the benzothiazole heterocycle will be introduced, highlighting how 2- aminobenzothiazole can serve as a scaffold conferring additional bioactivity to Schiff bases. By the end of this chapter, the specific **research problem** will be formulated: namely, the lack of comprehensive studies on Schiff bases derived from 2- aminobenzothiazole and ketones, and the potential value of examining their transition metal complexes. The **aim and objectives** of the present study will be clearly stated, articulating what the project seeks to accomplish and why it is important in the context of current knowledge.

Chapter 2: Literature Review – In this chapter, a thorough review of existing literature relevant to the topic will be presented. It will survey previous research on transition metal complexes of Schiff bases, with a focus on those involving benzothiazole or related N,S-heterocyclic ligands. For example, studies where 2-aminobenzothiazole was condensed with aromatic aldehydes (such as salicylaldehyde or substituted benzaldehydes) to create

Schiff base ligands will be discussed, since these are the closest precedents to the proposed ligands. The coordination modes observed in such complexes (typically bidentate N,N- chelation via the azomethine nitrogen and the benzothiazole ring nitrogen) and the geometries (often octahedral for divalent metal ions) will be summarized. Key findings from prior works – for instance, the enhanced **antimicrobial and antitumor activities** upon metal complexation of benzothiazole Schiff bases – will be reviewed in detail. Etaiw et al. (2011) observed that a 2-aminobenzothiazole Schiff base had relatively modest activity on its own, but its Cu(II), Ni(II), Zn(II) complexes showed significantly increased antimicrobial efficacy and cytotoxicity against tumor cell lines. Similarly, other studies on **DNA-binding** properties will be examined: Patil et al. (2022) demonstrated that certain Co(II), Ni(II), Cu(II) complexes of a benzothiazole-based ligand bind strongly to calf-thymus DNA, with Cu(II) showing the highest affinity as evidenced by spectroscopic titrations, viscosity measurements, and gel electrophoresis assays. Such literature evidence suggests that metallation can greatly influence the biological interactions of these ligands. The review will also encompass any known examples of Schiff bases derived from ketones (aliphatic or aromatic) to identify methodologies and outcomes relevant to our approach. Given that ketone-derived imines are somewhat less common than aldehyde-derived ones, any reported differences in stability or reactivity will be noted. Furthermore, very recent works that integrate computational chemistry in this domain will be highlighted – for instance, a 2025 study by Abd El-Nasser et al. which not only synthesized Ni(II), Cu(II), and Zn(II) complexes of a benzothiazole-containing ligand but also employed DFT calculations and molecular docking to explore their electronic structure and potential biomedical targets. By the end of the literature review, the **knowledge gap** that this Ph.D. project addresses will be clearly delineated: while benzothiazole Schiff base complexes are known to be promising, the subclass of aminobenzothiazole–ketone Schiff bases remains underexplored. This sets the stage for the subsequent chapters detailing our methodology and findings.

Chapter 3: Materials and Methods – This chapter will detail the experimental methodology adopted to achieve the research objectives. It will start with the **synthesis of the Schiff base ligands**: specifically, the condensation of 2-aminobenzothiazole with

selected ketone compounds to form the desired aminobenzothiazole–ketone Schiff bases. The procedure will likely involve refluxing the 2-aminobenzothiazole (which contains the primary amine group) with the ketone in an appropriate solvent (ethanol or methanol) in the presence of a trace acid catalyst to facilitate dehydration, if necessary. The choice of ketone substrates will be described (for example, acetylacetone for a bidentate ligand, or aromatic ketones like acetophenone or benzophenone derivatives for monodentate imines), along with rationales for their selection (different electronic or steric features to tune ligand properties). The isolation and purification of the Schiff base ligands (cooling-induced crystallization or recrystallization from solvents) will be outlined. Next, the **synthesis of metal complexes** will be presented. Transition metal salts of interest (such as Cu(II), Co(II), Ni(II), Zn(II), Fe(III), etc., typically as chlorides or acetates) will be reacted with the Schiff base ligands in roughly 1:2 metal-to-ligand molar ratio to yield the complexes. A general method to be used is mixing a hot ethanolic solution of the ligand with an ethanolic solution of the metal salt and refluxing the mixture for a certain period (e.g. 1 hour), during which the complex forms and often precipitates out. The resulting solid complexes will be filtered, washed with a suitable solvent (to remove unreacted ligand or salt), and dried. The chapter will then enumerate the **characterization techniques** employed for confirming structures and compositions of the synthesized compounds. These will include: Elemental analysis (CHN analysis) to verify the metal-to-ligand stoichiometry; Mass spectrometry for molecular weight confirmation; Fourier-transform infrared (FTIR) spectroscopy to identify key functional group vibrations. For example, the disappearance of the N–H stretching band of the 2-aminobenzothiazole (around ~3380 cm^−1) and the appearance of a strong azomethine C=N stretch near 1600 cm^−1 in the ligand will confirm successful Schiff base formation. Upon complexation, shifts in the ν(C=N) band (often to lower frequencies due to metal coordination) will indicate coordination through the imine nitrogen. Nuclear Magnetic Resonance (NMR) spectroscopy (^1H and ^13C, when applicable) will be used for the ligands and diamagnetic complexes (e.g. Zn(II) or Cd(II) complexes) to further confirm their structures (the azomethine proton signal around δ ~8–9 ppm typically shifts upon coordination). UV-Visible electronic spectroscopy will be conducted to observe ligand-field d–d transitions in the complexes and any π–π/n–π transitions of the ligands (these

data aid in assigning geometries and assessing electronic properties). Magnetic susceptibility measurements will be performed for paramagnetic complexes to determine their magnetic moments, which, in comparison with spin-only values, help deduce geometries and electron spin states (e.g. distinguishing between high-spin octahedral vs. low-spin or square-planar complexes). If single crystals of any complex can be obtained, X-ray crystallography will be attempted to definitively determine the coordination geometry and ligand denticity in the solid state. Additionally, the methods for evaluating **biological or catalytic activities** will be detailed here. For biological assays, the plan may involve antimicrobial tests (such as the disc diffusion method or MIC determination against select bacterial/fungal strains) to compare the efficacy of free ligands versus complexes. Any anticancer evaluation (e.g. an in vitro cell viability assay like MTT on cancer cell lines) will also be outlined. In the case of DNA-binding studies, protocols such as UV-Vis titration of complex with DNA, fluorescence quenching assays (with ethidium bromide bound DNA), viscosity measurements, and gel electrophoresis for DNA cleavage will be described. Finally, if computational techniques are used, the chapter will mention the use of theoretical calculations – for example, Density Functional Theory (DFT) to optimize structures or compute HOMO-LUMO gaps, and molecular docking simulations to predict how the complexes might interact with biomolecular targets (DNA or specific enzymes). By the end of Chapter 3, the reader will have a clear understanding of **how** the research will be carried out, in terms of both synthesis and analysis.

Chapter 4: Synthesis and Characterization of Aminobenzothiazole–Ketone Schiff Bases – This chapter will present the results obtained for the free Schiff base ligands (prior to metal complexation). It will typically begin by describing the physical appearance of each synthesized ligand (yield, color, melting point, solubility). Any interesting observation during synthesis (e.g. need for a catalyst, or formation of side-products like imidazolines in case of certain ketones) will be noted. The bulk of the chapter will then focus on spectroscopic characterization confirming the structures of these ligands. For each ligand, FTIR spectra will be discussed: the expected disappearance of the amino N–H bands from the starting 2-aminobenzothiazole and the appearance of the azomethine (C=N) stretch ~1600 cm^−1 provide clear evidence of

Schiff base formation. If the ketone carried any distinguishing functional groups (e.g. carbonyl in an acetylacetone may lead to enol- tethered imine, or aromatic substituents), their effects on the IR spectrum and ligand stability will be analyzed. NMR data (for protons and carbons) will be interpreted to assign the azomethine proton signal and other key signals; for instance, a singlet around δ ~8–9 ppm in ^1H NMR is characteristic of the –CH=N– proton, and its presence confirms the imine linkage. Comparisons will be made among the ligands if multiple ketones were used – discussing electron-withdrawing or donating substituent effects on the chemical shifts or infrared frequencies (e.g. how a ligand derived from an aromatic ketone with a nitro substituent might show a different C=N stretch frequency than one derived from an aliphatic ketone). UV-Vis spectra of the ligands may show π–π transitions of the benzothiazole ring and imine chromophore; any observable charge-transfer bands will be noted, as these can be relevant to their later complexation behavior. In addition, this chapter will report any unique properties of the free ligands*: for example, if any ligand exhibits fluorescence (some benzothiazole derivatives are fluorescent), or if preliminary antimicrobial tests indicate activity of the ligand alone. By thoroughly characterizing the Schiff bases in this chapter, a solid foundation is laid for understanding changes that occur upon metal coordination in the next chapter.

Chapter 5: Synthesis, Characterization, and Properties of Transition Metal Complexes

- This chapter constitutes the core of the research results, describing the obtained metal complexes of the Schiff base ligands and analyzing their properties. Initially, the chapter will detail the reaction outcomes for each metal: yields and colors of complexes, and any notable differences (for instance, complexes of the same ligand with different metals may have different colors or solubilities, hinting at differing structures or ligand-field strengths). The **stoichiometry** of each complex will be confirmed by elemental (CHN) analysis and, where possible, mass spectrometry, establishing formulas like $[M(L)_2Cl_2]$ or $[M(L)_2(anion)_n]$ as applicable. The discussion will then focus on spectroscopic evidence of coordination.

FTIR spectra of the complexes will be compared to those of the free ligands: typically, the azomethine ν(C=N) band shifts upon coordination (often to a lower wavenumber if

metal- to-ligand π-backbonding occurs, or to a higher wavenumber if the C=N bond order increases on coordination). For example, in similar benzothiazole Schiff base complexes, Co(II), Cu(II), Ni(II) coordination caused a lowering of the C=N stretch frequency due to back-donation from metal d-orbitals into the π of the imine. The presence or absence of other diagnostic bands will be noted: e.g. new bands in the far-IR that could indicate M–N or M–Cl bonds, or shifts in aromatic ring modes due to coordination. NMR spectroscopy (for diamagnetic complexes) will be used to see changes such as the deshielding or disappearance of the imine proton signal upon binding to a metal center (paramagnetic complexes will be analyzed by other means). Electronic (UV-Vis) spectra of the complexes will be presented, showing d–d transition bands characteristic of each metal's geometry (e.g. octahedral Cu(II) might show a broad band in the visible region). These, together with measured magnetic moments, will allow assignment of geometries: high-spin octahedral vs. tetrahedral vs. square-planar configurations will be distinguished by comparing to known standards. For instance, a Ni(II) complex might be found to be diamagnetic, suggesting a square-planar structure, whereas Co(II) and Cu(II) complexes are paramagnetic and likely octahedral. Any single-crystal X-ray structures obtained will be described here, providing definitive bond lengths and angles – this could reveal, for example, that the ligand L indeed chelates through the imine nitrogen and the benzothiazole ring nitrogen, forming five-membered chelate rings with the metal. In addition to structural characterization, this chapter will report on functional studies of the complexes. Results of antimicrobial assays will be given, comparing the efficacy of metal-bound ligands to the free ligands. It is expected (in line with published literature) that complexation often enhances biological activity; trends observed (such as one metal outperforming others against a particular microbe) will be discussed in terms of factors like increased lipophilicity of the complex aiding cell permeation. Any antioxidant or anticancer tests performed will also be presented, and the data (IC_50 values, etc.) interpreted. Furthermore, if DNA-binding experiments were conducted, their findings will be included – for example, UV-Vis titration graphs showing hypochromism or red-shift of the complex's absorbance upon adding DNA, or gel electrophoresis images indicating whether the metal complexes cleave DNA (perhaps in the presence of an oxidant). The binding strength (quantified by binding constants) and mode (intercalative

vs. groove-binding) will be inferred from these experiments. A comparison will be made among the series of complexes to see how changing the metal or the ketone substituents affects these properties. The chapter will also incorporate any relevant theoretical/computational results* if obtained: for instance, DFT-optimized structures might be shown to corroborate experimental geometry, and molecular docking simulations could illustrate how a representative complex fits into the DNA minor groove or interacts with a model protein. Any observed optical or electrochemical properties (e.g. if a complex shows interesting fluorescence or a redox activity in cyclic voltammetry) will be briefly noted as well. Overall, Chapter 5 will tie together the structural characterization with the functional evaluations, providing a comprehensive understanding of the synthesized transition metal complexes.

Chapter 6: Conclusion and Future Outlook – The final chapter will summarize the key findings of the research and present the conclusions drawn. It will succinctly reiterate how the **initial hypotheses** were addressed: for example, confirming that the aminobenzothiazole–ketone Schiff bases do act as effective bidentate ligands forming stable metal complexes, and that these complexes exhibit noteworthy properties (structural and biological) that justify the study. The chapter will discuss the significance of these findings in the broader context of coordination chemistry and potential real-world applications. Any trends observed across the series of complexes will be highlighted as conclusions – such as the relationship between ligand substituents and complex stability or activity, or the influence of metal ionic properties on the complex behavior. Importantly, this chapter will also identify **limitations** of the current work and suggest avenues for future research. For instance, if some complexes could not be crystallographically characterized, future work might focus on obtaining crystals or using advanced spectroscopic techniques (like EPR for Cu(II) complexes) to get more insights. If promising antimicrobial or anticancer activity was observed in vitro, the next step could be detailed mechanism-of-action studies or in vivo tests, which will be mentioned as future prospects. Additionally, the possibility of extending the chemistry to other metals or exploring catalytic reactions (such as oxidation or coupling reactions using these complexes as catalysts) will be proposed. By outlining these future directions, Chapter 6 will emphasize that the study of aminobenzothiazole–Schiff base metal complexes is a

fertile area for ongoing research, and the present work serves as a foundational step in that direction.

c) Hypothesis

- **Ligand Coordination**: The aminobenzothiazole–ketone Schiff base ligands are expected to function as bidentate chelators, coordinating to transition metal centers through the azomethine nitrogen and the benzothiazole ring nitrogen. This chelation will likely yield stable complex structures, such as six-coordinate octahedral complexes for divalent metals with a general formula [M(L)_2X_2] (where X = anion like Cl^–), or analogous four/five-coordinate structures depending on the metal and conditions. It is hypothesized that the presence of the benzothiazole ring (with its conjugated system and donor N atom) will enhance the ligand's binding affinity and possibly allow for interesting coordination modes (e.g. bridging between metals in polynuclear complexes or mixed N,S coordination if sulfur becomes involved under certain conditions).

- **Enhanced Properties upon Metallation**: It is further hypothesized that complexation with transition metals will **augment the functional properties** of the Schiff base ligands. In particular, the resulting metal complexes are expected to exhibit greater biological activity than the free ligands. This is based on the premise that many metal–Schiff base complexes show improved **antimicrobial and anticancer** effects relative to their uncoordinated ligands. The metal center can act synergistically with the ligand, for example by facilitating redox processes or improving cell uptake of the compound. Therefore, we anticipate that the synthesized complexes will demonstrate higher antibacterial/fungal efficacy and, if tested, higher cytotoxicity against cancer cells, compared to the ligand alone.

  Additionally, the complexes may show the ability to bind to biomolecules like DNA more strongly (especially certain metal ions such as Cu(II) which are known to promote DNA binding/cleavage) and could serve as artificial nuclease agents or enzyme inhibitors. These enhancements will be evidenced by spectroscopic and biological assays during the study.

d) Methodology

1. **Synthesis of Schiff Base Ligands**: Prepare a series of aminobenzothiazole–ketone Schiff base ligands by condensing 2-aminobenzothiazole with selected ketone compounds. This involves refluxing an ethanolic solution of 2-aminobenzothiazole with the ketone (in 1:1 molar ratio or slight excess of amine) to form the imine, with continuous removal of water. For less reactive ketones, use an acid catalyst (e.g. p- toluenesulfonic acid) or a Dean–Stark apparatus to facilitate dehydration. After reflux (several hours), cool the mixture to precipitate the Schiff base product, then filter and recrystallize it from a suitable solvent to obtain pure ligand. Record yields, melting points, and check purity (TLC or CHN analysis) for each ligand.

2. **Synthesis of Metal Complexes**: React each Schiff base ligand with various transition metal salts to form complexes. Typically, dissolve the ligand in hot ethanol and add an ethanolic solution of the metal chloride/acetate (metals to include Cu(II), Co(II), Ni(II), Zn(II), perhaps Fe(III) and others). Use a 2:1 ligand-to- metal molar ratio so that the metal can be fully chelated by two bidentate ligand molecules. Reflux the reaction mixture for ~1 hour, during which the complex often precipitates. Concentrate or cool the solution to ensure complete precipitation. Filter off the solid complex, wash with cold ethanol (to remove unreacted ligand or salt), and dry under vacuum. If the complex is oily or not precipitating, attempt precipitation by adding a non-polar solvent (diethyl ether) to the reaction mixture. **Repeat** this procedure for each metal–ligand combination in the study.

3. **Characterization of Ligands and Complexes**: Apply a comprehensive suite of analytical techniques to confirm structures:

4. Elemental Analysis (C, H, N): Determine the elemental composition of each complex, confirming the metal-to-ligand ratio and the presence of any anions or solvent of crystallization.
5. Mass Spectrometry: Obtain ESI-MS or MALDI spectra to identify molecular ion peaks corresponding to the [M(L)_2]^+ or related fragments, supporting the proposed formulas.
6. FTIR Spectroscopy: Record infrared spectra (4000–400 cm^−1) of each ligand and

complex. Note key changes: the appearance of the ν(C=N) imine band ~1600 cm^−1 in ligands (absent in the 2-aminobenzothiazole starting material), and shifts of this band in complexes (indicative of coordination). Identify other functional group bands (e.g. aromatic C=C, C–S–C of benzothiazole, M–N or M–Cl vibrations in far-IR if accessible).

7. NMR Spectroscopy: Acquire ^1H and ^13C NMR for all ligands and for diamagnetic complexes (e.g. Zn(II), Cd(II)). Verify the imine proton signal and its shift upon complexation. Use DMSO-d_6 or CDCl_3 as solvent depending on solubility.
8. UV-Vis Electronic Spectroscopy: Measure UV-Visible spectra of the complexes in solution (ethanol or DMSO). Identify ligand-centered π–π* transitions and any d–d bands. Use these to infer ligand field parameters and electronic transitions (e.g. estimate Δ_oct for octahedral complexes from d–d band positions).
9. Magnetic Susceptibility: Use a Gouy balance or SQUID magnetometer to measure the magnetic moment (μ_eff) of each complex at room temperature. Compare with spin-only values to determine electron configuration and geometry (e.g. μ_eff ~1.8–2.0 μ_B for one unpaired electron in Cu(II) indicates likely octahedral geometry, Ni(II) complexes with μ_eff ~0 (diamagnetic) would imply square-planar, etc.).
10. Single-Crystal XRD (if available): Attempt to grow single crystals of at least one representative complex by slow diffusion or evaporation (e.g. diffusing ether into a DMF solution). If successful, perform X-ray crystallography to obtain the exact molecular structure, bond lengths, and coordination geometry.
11. Thermal Analysis: Optionally, perform TGA/DTA on complexes to check for lattice solvents (weight loss) and thermal stability.
12. **Biological Activity Assays**: Evaluate the biological efficacy of the synthesized compounds:
13. Antimicrobial Tests: Use the agar diffusion method or broth microdilution to test each ligand and complex against representative bacterial strains (e.g. E. coli, S. aureus) and fungi (e.g. Candida albicans). Determine zones of inhibition or minimum inhibitory concentrations (MICs). Compare results to standard antibiotics and observe if complexes

show enhanced activity over free ligands, as often reported.

14. Cytotoxicity (Anticancer) Assay: If facilities permit, treat cultured cancer cell lines (such as HeLa, MCF-7, etc.) with varying concentrations of the complexes versus the free ligand. Use an MTT or SRB assay to measure cell viability and compute IC_50 values. This will reveal any increased cytotoxic effect due to metallation.
15. DNA-Binding/Cleavage Studies: Examine interaction with DNA using calf thymus DNA as a model. Conduct UV-Vis titration: gradually add DNA to a solution of the complex and monitor absorbance changes (hyperchromism or hypochromism, shifts in λ_max). Perform a viscosity experiment: measure the viscosity of a DNA solution with and without the complex (a classical intercalator will lengthen and thus increase viscosity, whereas a groove binder has lesser effect). Test nuclease activity by incubating the complex with plasmid DNA (pBR322, for example) in the presence of an oxidant and running agarose gel electrophoresis to see if DNA cleavage (nicked or linear forms) occurs. These experiments will shed light on the mode and strength of DNA interaction, an important aspect for potential anticancer mechanisms.
16. Enzyme Inhibition Assays: If relevant to the hypothesis, screen the complexes against certain enzymes (for example, urease or tyrosinase, if the ligand design suggests it) to see if they act as inhibitors, which could add pharmacological value.

All experimental procedures will be conducted following standard laboratory safety and quality protocols. Data will be recorded and analyzed and each set of results will be cross-validated (for example, confirming metal content by both elemental analysis and gravimetric oxide yield if needed). This multi-pronged methodological approach is designed to thoroughly investigate both the **coordination chemistry** and the **potential applications** of the new aminobenzothiazole–ketone Schiff base complexes.

e) Bibliography

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